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1.	Your reference	4-32797P1
2.	Patent application number *(The Patent Office will fill in this part)*	0229020.3 12 DEC 2002
3.	Full name, address and postcode of the or of each applicant *(underline all surnames)*	NOVARTIS AG LICHTSTRASSE 35 4056 BASEL SWITZERLAND
	Patent ADP number *(if you know it)*	7125487005.
	If the applicant is a corporate body, give the country/state of its incorporation	SWITZERLAND
4.	Title of invention	Organic compounds
5.	Name of your agent *(If you have one)*	
	"Address for service" in the United Kingdom to which all correspondence should be sent *(including the postcode)*	B.A. YORKE & CO. CHARTERED PATENT AGENTS COOMB HOUSE, 7 ST JOHN'S ROAD ISLEWORTH MIDDLESEX TW7 6NL
	Patents ADP number *(if you know it)*	1800001
6.	If you are declaring priority from one ore more earlier patent applications, give the country and the date of filing of the or of each of these earlier applications and *(if you know it)* the or each application number	Country / Priority application number *(if you know it)*
7.	If this application is divided or otherwise derived from an earlier UK application, give the number and the filing date of the earlier application	Number of earlier application / Date of filing (day/month/year)
8.	Is a statement of inventorship and of right to grant of a patent required in support of this request? *(Answer 'Yes' if:* *a) any applicant named in part 3 is not an inventor, or* *b) there is an inventor who is not named as an applicant, or* *c) any named applicant is a corporate body.* *(see note (d))*	Yes

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Claim(s)	3
Abstract	
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Statement of inventorship and right to grant of a patent *(Patents Form 7/77)*	
Request for preliminary examination and search *(Patents Form 9/77)*	One
Request for substantive examination *(Patents Form 10/77)*	
Any other documents *(please specify)*	

11. I/We request the grant of a patent on the basis of this application

Signature Date

B.A. Yorke & Co. 12 December 2002

12. Name and daytime telephone number of person to contact in the United Kingdom

Mrs. J. Crook
020 8560 5847

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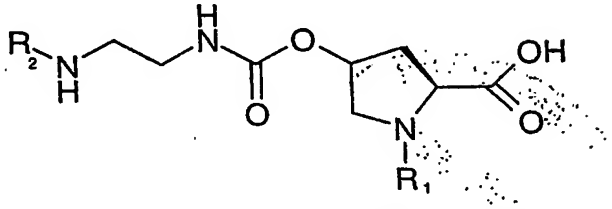
Organic compounds

The present invention relates to a process for producing organic compounds, and to intermediates produced in such a process.

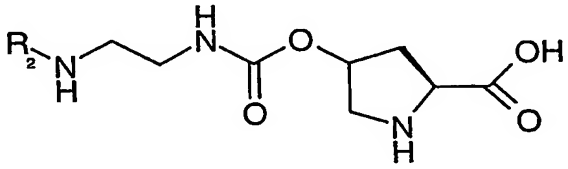
More particularly, the invention relates to:

(A) a process for preparing a compound of formula I



I

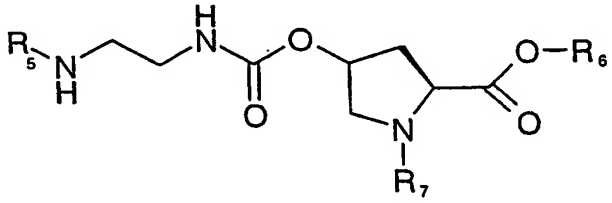
wherein R_1 and R_2 are each a removable protecting group and R_1 and R_2 are different; comprising reacting a compound of formula II



II

with a suitable R_1 donor compound;

(B) intermediates useful in the above process, defined by the general formula VII



VII

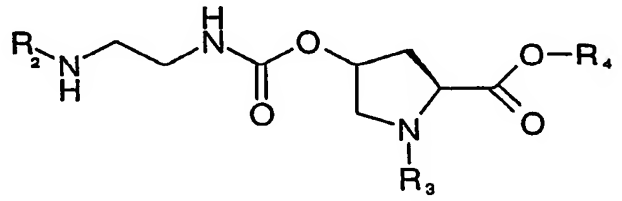
wherein R_5 is a removable protecting group other than fluorenylmethoxycarbonyl, and is different to R_7;

R_6 is hydrogen or a blocking group removable by hydrolysis or hydrogenolysis; and

R_7 is hydrogen or a removable protecting group other than fluorenylmethoxycarbonyl.

The present invention provides a simple and efficient route for the preparation of compounds of formula I, which are useful in the synthesis of peptides, for example as described in WO 02/10192. The compounds of formula VII are useful as intermediate compounds in the preparation of compounds of formula I.

The compound of formula II may be prepared from a compound of formula III



III

wherein R_2 is as defined above,
R_3 is a removable protecting group and R_3 is different to R_1 and R_2, and
R_4 is a blocking group removable by hydrolysis or hydrogenolysis.

Protecting groups, their introduction and removal are described, for example, in "Protective Groups in Organic Synthesis", T. W. Greene et al., John Wiley & Sons Inc., Second Edition 1991. Suitable protecting group donor compounds, e.g. amino group protecting agents, are well-known to a skilled person, e.g. anhydrides, halides, carbamates or N-hydroxysuccinimides which provide one of the protecting groups below.

The protecting group R_1 is preferably fluorenylmethoxycarbonyl. R_2 or R_5 is preferably a protecting group other than fluorenylmethoxycarbonyl, and is preferably more resistant to removal by hydrolysis (for example base-catalysed hydrolysis) and/or hydrogenolysis than R_1 and/or R_3, e.g. more resistant than fluorenylmethoxycarbonyl and/or benzyloxycarbonyl. More preferably R_2 or R_5 is tert-butoxycarbonyl.

The protecting group R_3 or R_7 is preferably more resistant to removal by hydrolysis than R_1, e.g. more resistant than fluorenylmethoxycarbonyl. R_3 or R_7 is preferably removable by hydrogenolysis. Suitable R_3 or R_7 substituents include benzyloxycarbonyl, 1,1,-dimethylpropynyloxycarbonyl, vinyloxycarbonyl, N-hydroxypiperidinyloxycarbonyl, 9-anthrylmethyloxycarbonyl and phenylaminothiocarbonyl, allyl, nitrobenzyl, triphenylmethyl, (p-methoxyphenyl)diphenylmethyl, diphenyl-4-pyridylmethyl or benzylsulfonyl. Preferably R_3 or R_7 is an oxycarbonyl-containing protecting group, e.g. benzyloxycarbonyl (carbobenzoxy).

R_4 or R_6 may suitably be:

(i) C_{1-10}-alkyl, e.g. C_{1-4}-alkyl, preferably methyl, ethyl, propyl or butyl other than tert-butyl, more preferably methyl.

(ii) C_{3-8}-cycloalkyl, optionally substituted by one or more C_{1-4} alkyl, e.g. methyl. Preferably cycloalkyl is C_{3-6}-cycloalkyl.

(iii) C_{6-10}-aryl, optionally substituted by one or more stabilising substitutents, e.g halogeno or nitro. Preferably aryl is phenyl, optionally substituted by one, two or three halogeno, e.g. chloro.

(iv) $(C_{6-10}\text{-aryl})_{1-3}$-$C_{1-10}$-alkyl, optionally substituted on the aryl group by (i) one or more stabilising substituents, e.g halogeno or nitro, or (ii) by two substituents which together with the ring carbon atoms to which they are attached form a 5- or 6-membered ring, optionally containing one or two nitrogen or oxygen atoms. $(C_{6-10}\text{-aryl})_{1-3}$-$C_{1-10}$-alkyl is preferably (i) $(\text{phenyl})_{1-3}$-C_{1-4}-alkyl, more preferably benzyl, diphenylmethyl or triphenylmethyl, optionally substituted on each benzene ring by one, two or three halogeno, e.g chloro, (ii) anthrylmethyl, e.g. 9-anthrylmethyl, or (iii) piperonyl.

(v) C_{6-10}-aryl-C_{1-4}-alkoxy-C_{1-4}-alkyl, preferably benzyloxymethyl.

(vi) C_{6-10}-aryl-carbonyl-C_{1-4}-alkyl, preferably phenacyl.

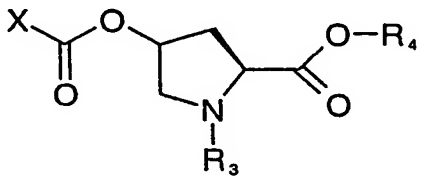
In the above, alkyl means straight or branched alkyl. Preferably R_4 or R_6 is a group which is removable by hydrogenolysis, such as benzyl, benzyloxymethyl, phenacyl, triphenylmethyl, piperonyl or 9-anthrylmethyl, preferably benzyl.

The compound of formula II may be prepared by (i) hydrolysing the ester compound of formula III to obtain the corresponding carboxylic acid and (ii) removing the protecting group R_3. Preferably the hydrolysis step is performed before removal of the protecting group R_3. The protecting group R_3 may conveniently be removed by reductive hydrogenation (hydrogenolysis). This route, involving a hydrolysis step, is suitably followed when R_4 is not removable by hydrogenolysis. The hydrolysis step is preferably a base-catalysed hydrolysis,

for example using sodium hydroxide and may suitably be performed in a polar solvent, e.g.methanol.

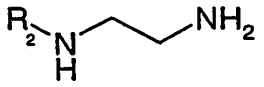
Alternatively, a compound of formula II may conveniently be prepared by hydrogenation (hydrogenolysis) of a compound of formula III wherein R_4 is a group which is removable by hydrogenolysis, e.g. benzyl. The hydrogenation step may conveniently be performed using a suitable catalytic agent, for instance palladium-on-charcoal.

Compound of formula III may be prepared by reacting a compound of formula IV



IV

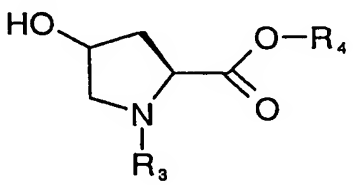
wherein X is a nucleophilic substituent and R_3 and R_4 are as defined above, with a compound of formula V



V

wherein R_2 is as defined above. This step may be performed in any suitable organic solvent, preferably in a hydrocarbon solvent, more preferably toluene.

The compound of formula V is a protected ethylenediamine (diaminoethane), wherein one amino group has been protected with a removable protecting group. The nucleophilic substituent X in formula IV is preferably halogeno, such as fluoro, chloro, bromo or iodo, more preferably chloro. The compound of formula IV wherein X is halogeno may be formed by reaction of a compound of formula VI



VI

with an acyl halide, for instance phosgene, tri-phosgene, phenylchloroformate or 4-nitrophenylchloroformate, preferably 4-nitrophenylchloroformate. This step may suitably be performed in the presence of an organic base, e.g. dimethylaminopyridine, in a non-polar solvent, e.g. toluene.

The compound of formula VI may be commercially available, e.g. when R_4 is methyl or may be formed by esterification of 4-hydroxy-proline according to methods known in the art, for instance by reaction with benzyl alcohol or methanol. The resulting ester is then protected by reaction with a suitable R_3 donor compound, e.g. benzyloxycarbonyl-N-hydroxysuccinimide.

The compound of formula IV need not be separated or isolated, as the compound of formula VI may be reacted with an acyl halide and the product of this reaction subsequently reacted with a compound of formula V in the same vessel.

The addition of the protecting group R_1 to the compound of formula II may suitably be performed in the presence of sodium carbonate/acetonitrile.

Compounds of formula I can be recovered from the reaction mixture and purified in a conventional manner.

In the compounds of formulae I-IV and VI above, the oxy substituent on the proline may be in position cis or trans, preferably trans. The cis or trans isomers may be individually prepared, using the corresponding cis or trans hydroxyproline as starting material.

Insofar as the production of the starting materials is not particularly described, the compounds are known or may be prepared analogously to methods known in the art or as described thereafter.

In a further aspect, the present invention relates to a process for producing a compound of formula I, wherein R_1 is fluorenylmethoxycarbonyl and R_2 is a removable protecting group other than fluorenylmethoxycarbonyl, comprising reacting a compound of formula II with a fluorenylmethoxycarbonyl donor compound, e.g. fluorenylmethoxycarbonyl-N-hydroxysuccinimide.

The invention will now be described with reference to the following specific embodiments, in which the following abbreviations are used:

Fmoc = fluorenylmethoxycarbonyl
Boc = tert-butoxycarbonyl
Cbo = carbobenzoxy (benzyloxycarbonyl)
OSu = N-hydroxysuccinimide

Example 1

Preparation of Fmoc-(2S,4R)-Pro(4-OCO-NH-CH_2-CH_2-NH-Boc)-OH starting from Cbo-(2S,4R)-Pro(4-OH)-OMe

1. Dimethylaminopyridine (30.5 g, 250 mmol) and Cbo-(2S,4R)-Pro(4-OH)-OMe (34.9 g, 125 mmol) are dissolved in toluene (870 ml). A solution of 4-nitrophenylchloroformate (31.5 g, 157 mmol) in toluene (206 ml) is added dropwise to this solution at 0°C to 5 °C over 20 minutes and stirred for an additional 2 hours. This is followed by addition of a solution of Boc-ethylenediamine (80.1 g, 500 mmol) in toluene (205 ml) and stirring at ambient temperature for 12 hours. A solution of concentrated sulfuric acid (43.7 g, 450 mmol) in water (873 ml) is then added while maintaining a temperature of 20 °C to 25 °C. The white suspension is filtered by suction and washed with toluene (30 ml). The toluene phase is washed with water (450 ml), sodium carbonate (10% w/w, 450 ml) and three times with water (450 ml each). The toluene phase is azeotropically dried by distilling off 300 ml, which is continuously replaced by dry toluene (2 x 300 ml). Heptane (130 ml) is added to the dry toluene solution at 50 °C and cooled to 0°C over two hours. The precipitated product is filtered, washed two times with toluene/heptane 1:2 v/v (70 ml), and dried at 50 °C under vacuum to leave Cbo-(2S,4R)-Pro(4-OCO-NH-CH_2-CH_2-NH-Boc)-OMe as a white solid.

2. Cbo-(2S,4R)-Pro(4-OCO-NH-CH_2-CH_2-NH-Boc)-OMe (20.0 g, 43.0 mmol) is dissolved in a 1:1 mixture of tetrahydrofuran and methanol (380 ml). A 1 M sodium hydroxide solution (51.6 ml) is added and the resulting mixture stirred for 4 hours at ambient temperature. The mixture is adjusted to pH 3 by adding sulfuric acid (50 ml, 1 M). Tetrahydrofuran and methanol are distilled off at 50 °C and 50 mbar until no further solvents distil. The remaining milky solution is diluted with isopropyl acetate (113 ml) and water (57 ml), the phases are separated and the isopropyl acetate phase is washed with sodium chloride solution (10%, 113 ml). The solvent is distilled off (50 °C, 50 mbar) to yield a foam of Cbo-(2S,4R)-Pro(4-

OCO-NH-CH_2-CH_2-NH-Boc)-OH (19.8 g), which was used without further purification in the next reaction.

3. Palladium on charcoal (10%, 1.94 g, 0042 mmol) is added to a solution of Cbo-(2S,4R)-Pro(4-OCO-NH-CH_2-CH_2-NH-Boc)-OH (19.4 g, 43.0 mmol) in isopropanol (350 ml) and water (37 ml). Hydrogen is bubbled through this mixture for 4 hours, the catalyst is filtered off, and the residue is washed with a mixture of isopropanol (50ml) and water (50 ml). The isopropanol/ water phase is azeotropically dried by distilling off 2/3 of the volume, which is continuously replaced by a toluene/isopropanol mixture (1:1 v/v). The remaining dry solution is concentrated *in vacuo* to dryness (50 °C, 200 mbar) to leave (2S,4R)-Pro(4-OCO-NH-CH_2-CH_2-NH-Boc)-OH as a brownish solid, which was used without further purification.

4. (2S,4R)-Pro(4-OCO-NH-CH_2-CH_2-NH-Boc)-OH (5.0 g, 15 mmol) is dissolved in a mixture of water (25 ml) and triethylamine (1.5 g, 15 mmol) at 40 °C. A solution of Fmoc-OSu (4.65 g, 14 mmol) in acetonitrile (25 ml) is added to the clear solution over 30 minutes and stirred for 2 hours. Then the reaction mixture is adjusted to pH 3 with hydrochloric acid (1 m, 13 ml) and stirred for a further hour. Acetonitrile is distilled off (40 °C, 80 mbar) and replaced by isopropyl acetate, affording a two-phase mixture. The lower aqueous phase is separated off, whilst the remaining organic layer is washed with water and distilled two times with replacement with isopropylacetate and then concentrated to a brownish foam. This foam is dissolved in isopropylacetate (25 ml) and added dropwise to heptane (200 ml) whereby the product is precipitated. The solid is filtered, washed with isopropylacetate/heptane and dried *in vacuo* at 40 °C to leave Fmoc-(2S,4R)-Pro(4-OCO-NH-CH_2-CH_2-NH-Boc)-OH.

Example 2

Preparation of Fmoc-(2S,4R)-Pro(4-OCO-NH-CH_2-CH_2-NH-Boc)-OH starting from Cbo-(2S,4R)-Pro(4-OH)-OBzl

The synthesis of Cbo-(2S,4R)-Pro(4-OH)-OBzl is described in T. Makoto, H. Guoxia, V. J. Hruby, J. Org. Chem. 2001, 66, 1038-1042. The process of example 1 is repeated, but using Cbo-(2S,4R)-Pro(4-OH)-OBzl in place of Cbo-(2S,4R)-Pro(4-OH)-OMe and performing steps 1, 3 and 4 only (omitting step 2).

Example 3

Preparation of Fmoc-(2R,4R)-Pro(4-OCO-NH-CH_2-CH_2-NH-Boc)-OH

The process of example 1 or example 2 is repeated but using Cbo-(2R,4R)-Pro(4-OH)-OMe or Cbo-(2R,4R)-Pro(4-OH)-OBzl in place of Cbo-(2S,4R)-Pro(4-OH)-OMe or Cbo-(2S,4R)-Pro(4-OH)-OBzl.

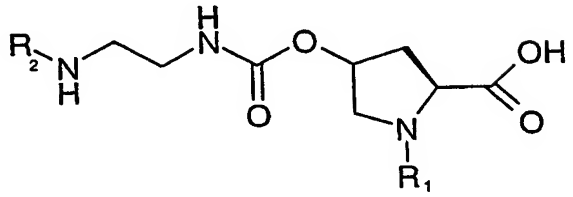
Example 4

Preparation of Fmoc-(2S,4S)-Pro(4-OCO-NH-CH_2-CH_2-NH-Boc)-OH

The process of example 1 or example 2 is repeated but using Cbo-(2S,4S)-Pro(4-OH)-OMe or Cbo-(2S,4S)-Pro(4-OH)-OBzl in place of Cbo-(2S,4R)-Pro(4-OH)-OMe or Cbo-(2S,4R)-Pro(4-OH)-OBzl.

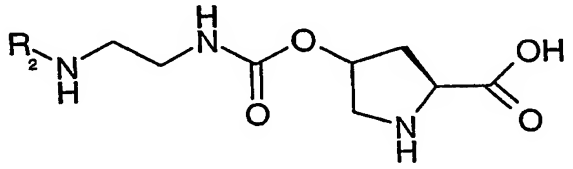
Claims

1. A process for preparing a compound of formula I



I

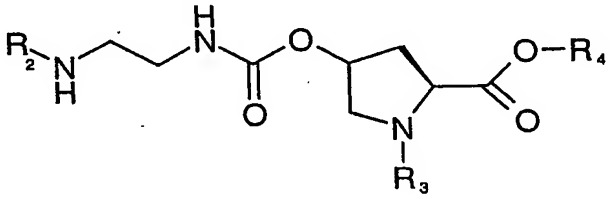
wherein R_1 and R_2 are each a removable protecting group and R_1 and R_2 are different; comprising reacting a compound of formula II



II

with a suitable R_1 donor compound.

2. A process according to claim 1, wherein the compound of formula II is prepared by (i) hydrolysing a compound of formula III

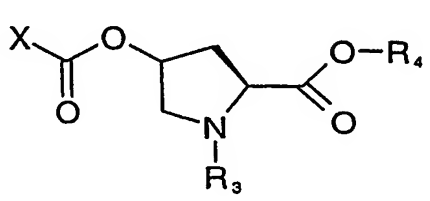


III

wherein R_2 is as defined in claim 1,
R_3 is a removable protecting group and R_3 is different to R_1 and R_2, and
R_4 is a blocking group removable by hydrolysis or hydrogenolysis,
to obtain the corresponding carboxylic acid, and
(ii) removing the protecting group R_3 in the resulting carboxylic acid.

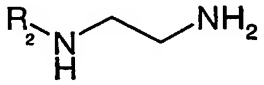
3. A process according to claim 1, wherein the compound of formula II is prepared by hydrogenating a compound of formula III, wherein each of R_3 and R_4 is a group removable by hydrogenolysis.

4. A process according to claim 2 or claim 3, wherein the compound of formula III is prepared by reacting a compound of formula IV



IV

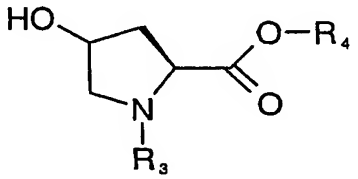
wherein X is a nucleophilic substituent
and R_3 and R_4 are as defined in claim 2 or claim 3,
with a compound of formula V



V

wherein R_2 is as defined in claim 1.

5. A process according to claim 4, wherein the compound of formula IV is prepared by reaction of a compound of formula VI



VI

wherein R_3 and R_4 are as defined in claim 2 or claim 3,
with an acyl halide.

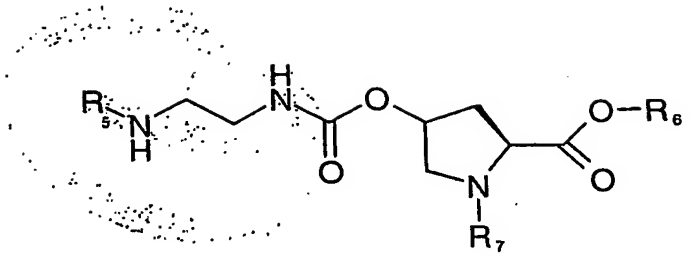
6. A process according to any of claims 2 to 5, wherein R_4 is methyl, ethyl, propyl, butyl other than tert-butyl, benzyl, benzyloxymethyl, phenacyl, triphenylmethyl, piperonyl or 9-anthrylmethyl.

7. A process according to any preceding claim, wherein R_2 is tert-butoxycarbonyl.

8. A process according to any preceding claim, wherein R_1 is fluorenylmethoxycarbonyl.

9. A process according to any preceding claim, wherein R_3 is benzyloxycarbonyl.

10. A compound of formula VII



VII

wherein R_5 is a removable protecting group other than fluorenylmethoxycarbonyl, and is different to R_7;
R_6 is hydrogen or a blocking group removable by hydrolysis or hydrogenolysis; and
R_7 is hydrogen or a removable protecting group other than fluorenylmethoxycarbonyl.

13. A compound according to claim 12, wherein R_5 is tert-butoxycarbonyl.

14. A compound according to claim 12 or claim 13, wherein R_7 is hydrogen or a protecting group more resistant to base-catalysed hydrolysis than fluorenylmethoxycarbonyl.

15. A compound according to claim 14, wherein R_7 is benzyloxycarbonyl.

16. A compound according to any of claims 12 to 15, wherein R_6 is hydrogen or methyl.

17. A compound according to any of claims 12 to 15, wherein R_6 is removable by hydrogenolysis.

18. A compound according to claim 17, wherein R_6 is benzyl.

19. A process for producing a compound of formula I, wherein R_1 is fluorenylmethoxycarbonyl and R_2 is a removable protecting group other than fluorenylmethoxycarbonyl, comprising reacting a compound of formula II with a fluorenylmethoxycarbonyl donor compound.

20. A process substantially as hereinbefore described with reference to the examples.

21. An intermediate compound, excluding starting materials and product, substantially as hereinbefore described with reference to the examples.